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1.0 Executive Summary

The Neonicotinoid Consortium is made up of the four primary registrants (Bayer Crop Science, Syngenta Crop Protection LLC, Valent U.S.A. LLC and Mitsui Chemicals Agro, Inc.) of the four nitroguanidine neonicotinoid pesticides (imidacloprid, thiamethoxam, clothianidin and dinotefuran). The Consortium was formed to serve as a vehicle for collaboration on the development of protocols and generation of data to support the nitroguanidine class of neonicotinoid insecticides under registration review.

The Neonicotinoid Consortium strongly supports the EPA and their efforts in providing a science-based risk assessment process for the nitroguanidine neonicotinoid insecticides which represents a culmination of effort to advance the bee risk assessment process as outlined in the Guidance for Assessing Pesticide Risk to Bees (EPA 2014). While the preliminary bee risk assessments for the nitroguanidine neonicotinoid insecticides, in general, follows a science-based risk assessment process outlined in EPA's guideline, the Neonicotinoid Consortium does have significant concerns about some of methods used in the preliminary risk assessments that were outside of the methods proposed in EPA's bee risk assessment guidance.

The Neonicotinoid Consortium comments are divided into the following topics which contain both criticisms (e.g., bee bread) and support (e.g., non-Apis assessment) for the Agency-proposed methodology and, where appropriate, recommendations on alternative approaches. These recommendations are provided based on the understanding that the methodologies presented in the preliminary bee risk assessments are not final given that they were not specifically detailed in the bee risk assessment guidance document and that there is opportunity to make appropriate changes to these proposed methodologies prior to the release of the final risk assessments.

The Agency evaluations of the colony feeding studies. These studies represent the best available science and greatly inform the pollinator risk assessments at the Tier II level especially with respect to the effects of chronic exposure to honey bee colonies. Overall, the Neonicotinoid Consortium agrees with the Agency on the use and general interpretations of the colony feeding studies to date and their use as higher-tier studies (Tier II) in the preliminary bee risk assessments. However, comments provided by the Agency in the preliminary assessments and Data Evaluations Records (DERs) for the colony feeding studies are questionable and

deserve further scrutiny, including analysis of certain endpoints lacking biological significance, seasonal timing of exposure, interpretation of the required feeding volumes, and lack of understanding of the normal control colony performance. The seasonal timing of exposure for all of the colony feeding studies were very similar as the Agency requested that the studies start during the natural nectar dearth period for the geography of the study (late June or early July for NC). In addition, the data show that control colonies behaved similarly in all four colony feeding studies and that this pattern of growth is typical for honey bee colonies in the Southeast U.S. Based on the poor overwintering survivorship in the controls, the EPA had asked the Registrants for clothianidin and thiamethoxam to repeat the colony feeding studies in 2016. Given that both studies were able to assess potential effects to colonies prior to and post winter (and confirmed effects observed in the previous studies), the Neonicotinoid Consortium recommends that the 2016 studies for these compounds be used quantitatively in the updated final ecological risk assessment.

Use of bee bread exposure in the risk assessment. The Neonicotinoid Consortium has significant concerns with the use of EPA's bee bread calculations in determining risk to bees. The formula used appears to have an error that overestimates bee bread concentrations. Furthermore, the approach of defining a toxicity endpoint based on the concentration in bee bread of the lowest-observed-effect level of the colony-feeding study assumes the response of the colony is driven by exposure through bee bread, when this represents only 0 to 27% (average 11%) of the total dose received by the various castes of worker bees in the colony. There is also empirical evidence (previous work with imidacloprid) that confirms that exposure from ingestion of residues in nectar, not pollen or bee-bread, drives the response of honey bees to neonicotinoids. For Tier II risk assessment, it is recommended that field measurements of residue levels in pollen be converted to nectar equivalents so that the total dietary concentration (pollen and nectar combined) can be directly compared to the NOAEC and LOAEC of the existing colony-feeding studies.

Risk Evaluation to non-*Apis* Bees. The Neonicotinoid Consortium supports the Agency's comprehensive weight of evidence analysis for evaluating risks to non-*Apis* species. However, the ability to reliably determine a no effect-concentration in non-*Apis* species or estimate their exposure levels in a quantitative basis is currently limited. Work in both exposure and effects aspects of the risk assessment for non-*Apis* bees is progressing, but further research is needed. In the interim, the Neonicotinoid Consortium agrees with the EPA that there is sufficient body of evidence for the nitroguanidine neonicotinoids suggesting that honey bees are reasonable surrogates.

Off-field risk assessment. EPA used the AgDRIFT model to estimate the fraction of the foliar-applied application rate at various distances beyond the treated field. The off-field risk conclusions, which are based on overly conservative and rather simplistic assumptions concerning drift, could potentially impact the use of any foliar spray applications regardless of crop attractiveness to pollinators or agronomic practices. The Neonicotinoid Consortium recommends that the Agency consider refinements to the AgDRIFT model when supported by label language to provide a more realistic estimate of potential exposure considering the drift deposition estimates are highly conservative. If available, drift deposition data from field drift

studies with formulated products of the AI should be used in place of AgDRIFT estimates. In addition, if available, No Observable Effect application Rates (NOERs) from semi-field tunnel studies should be used to compare rates to the AgDRIFT deposition curve to identify distances appropriate for protecting honey bee colonies. If semi-field data are not available, the acute contact LD50 should be used in conjunction with BeeRex exposure values determined from drift deposition estimates to calculate RQs that are compared to the acute LOC. Acute oral and chronic oral risk components are not necessary as the potential area of forage that would receive drift deposition would be small compared to the forage range of honey bees and drift deposition onto pollen and nectar would be low such that potential risk from oral exposure would be minimal.

Use of Residue Data in the Exposure Assessment. The Neonicotinoid Consortium have submitted to the Agency an extensive data set of crop residue data (nectar and pollen) developed to derive bee exposure estimates. In the refined Tier I assessment, EPA's approach is to use the overall maximum values and the maximum mean values for individual sites to derive estimated environmental concentrations (EECs). The Neonicotinoid Consortium believes that this approach is overly conservative as many of the maximum values are outliers and do not represent the majority of the data and that better use of the full data set could be made to derive a more realistic exposure estimate. The Consortium recommends that for acute risk, 90th percentile residue values can be derived from these data sets while for the chronic risk, overall mean values from all sites can be used.

Seed Treatment Dust Stewardship. The Neonicotinoid Consortium recognizes that exposure of bees via drift of abraded seed coat dust is considered a route of concern by the Agency and that "the Agency is working with different stakeholders to identify best management practices and to promote technology-based solutions that reduce this potential route of exposure." We provide some examples of stewardship efforts in this regard including ongoing efforts to develop and optimize new seed treatment formulations and tank-mix recipes to minimize dust abrasion through the use of new and improved dust reducing agents and polymers.

2.0 Introduction

The Neonicotinoid Consortium is made up of the four primary registrants (Bayer Crop Science, Syngenta, Valent and Mitsui Chemicals Agro, Inc.) of the four nitroguanidine neonicotinoid pesticides (imidacloprid, thiamethoxam, clothianidin and dinotefuran). The Consortium was formed to serve as a vehicle to collaborate on the development and harmonization of protocols used to generate data required by the Agency to perform pollinator risk assessments of the nitroguanidine class of neonicotinoid insecticides under registration review.

The United States Environmental Protection Agency (EPA) recently released preliminary bee risk assessments as part of the registration review of clothianidin and thiamethoxam as a single risk assessment (EPA-HQ-OPP-2011-0581-0034) and dinotefuran (EPA-HQ-OPP-2011-0920-0014) and also an update to its preliminary risk assessment for imidacloprid (EPA-HQ-OPP-2008-0844-0140). The dinotefuran assessment represents a screening level assessment, including refinement from use of pollen and nectar residue data collected from variety of crops. The

preliminary bee risk assessment for clothianidin and thiamethoxam and imidacloprid include a higher-tier, refined assessment that incorporates data and information currently available to the Agency including data on pollen and nectar residues and higher-tier effects data (i.e., colony feeding studies). Additional data for the nitroguanidine neonicotinoids have been collected and compiled since the release of the assessment and will be incorporated in an updated ecological risk assessment. It is anticipated that a higher-tier, refined assessment for dinotefuran will be performed similarly to the other compounds. The EPA has asked for public comments on the preliminary bee risk assessment including the bee bread methodology which represents a novel approach that has not been used previously in any assessment including the imidacloprid preliminary bee risk assessment that was released in January 2016 (EPA-HQ-OPP-2008-0844-0140).

The Neonicotinoid Consortium strongly supports the EPA and their efforts in providing a science-based risk assessment process for the nitroguanidine neonicotinoid insecticides which represent a culmination of effort to advance the bee risk assessment process as outlined in the Guidance for Assessing Pesticide Risk to Bees (USEPA, PMRA, CDPR 2014)[1]. While the preliminary bee risk assessments, in general, follow the science-based risk assessment process outlined in EPA's guideline, the Neonicotinoid Consortium does have significant concerns about some of methods used outside of the methods proposed in EPA's bee risk assessment guidance.

3.0 Colony Feeding Study Comments

In several places in the Data Evaluation Record (DER) for the clothianidin (EPA-HQ-OPP-2011-0865-0179) and the thiamethoxam (EPA-HQ-OPP-2011-0581-0040) colony feeding studies, the Agency expresses uncertainty in the control performance. Contrary to this conclusion, the consortium believes that the increased replication for control colonies, multiple pre-treatment assessments, and independent apiary locations result in a robust study design for assessing control performance. If the Agency believes that current assessment methodologies are insufficient for evaluating the performance of honey bee colonies, then this represents a very significant problem with current study designs which should be addressed.

In any toxicology study, the control or reference group is the most critical for accurate interpretation of any findings. Simply put, one cannot characterize what is abnormal due to treatment if one does not understand what is normal. Inherent in the study design for these colony feeding studies are measures meant to establish a high quality baseline with which to compare to the various treatment concentrations.

Multiple colony condition assessments are performed before the treatment begins in order to provide data on the quality of the test colonies. As those colonies in the chemical treatment groups have yet to be exposed, there is a wealth of data with which to understand the starting conditions of these experiments. In the DERs, the Agency states "The fact that many of the hives in the lower treatment

[1] USEPA, PMRA and CDPR (2014) Guidance for Assessing Pesticide Risks to Bees. Office of Pesticide Programs, United States Environmental Protection Agency, Washington, D.C.; Health Canada Pest Management Regulatory Agency Ottawa, ON, Canada California Department of Pesticide Regulation, Sacramento, CA. June 19. (available at: http://www2.epa.gov/pollinatorprotection/pollinator-risk-assessment-guidance).

groups performed/trended similarly to the control hives for these measurements could be indicative of either a lack of treatment effects or potentially/simply that the *control hives were suboptimal to begin the study*." Considering the multiple assessments and endpoints collected from colonies before initiation of the treatments in these studies, suboptimal control colonies should be identifiable and removed from the study. If the conducted measurements are not considered effective by the Agency for evaluating the condition or performance of colonies, this should be communicated by the Agency for future studies.

Similarly, it is stated of the control colonies in both clothianidin and thiamethoxam DERs that "it is still uncertain if the hives were developing normally". Based on the data collected on multiple endpoints at multiple assessments, if control hive performance was not being characterized adequately to evaluate if control hives were normally developing, this should have been indicated by the Agency.

In the thiamethoxam colony feeding study DER, it is stated "since no sustained significant effects were seen except in the 100 µg/L treatment it is uncertain if the control hives were performing normally during the test." This sentence as written is particularly troubling to the Consortium as it implies that the observations of statistical differences between control and treatment groups are being used to determine if control hives perform normally. Control hive performance should be evaluated completely independently of any observed statistical difference between the control and any other treatment group. An observation of statistical differences between the control and a treatment group does not increase certainty that control hives are performing normally.

The Agency concludes that it is likely that the provided amount of treatment sucrose solutions during the colony feeding study does not meet the carbohydrate needs of the colonies. However, based on published estimates of colony consumption of honey during the summer period, feeding rates appear to not only meet, but exceed the dietary needs of the colonies. This is further supported by measurements taken during the study such as increasing honey and nectar food stores.

In the DER for the clothianidin colony feeding study, it is stated: "The quantity of nectar provided to hives (4 L per week per hive) likely did not fulfill the complete carbohydrate needs of the colony, as indicated by colony bioenergetics and the lack of remaining sucrose solution upon their renewal at some of the test concentrations." Colonies in this study were provided 4 L of 50% sucrose solution every week. Assuming a density of 1.23 g/cm^3, this volume would be 4.92 kg added each week. Assuming honey is 80% sugar, this 4.92 kg of 50% sucrose solution when dried down to a honey-like level would weigh 3.07 kg. Therefore the amount of feeding of treatment solutions on a weekly basis is equivalent to 3.07 kg of honey.

In: Honeybee Ecology: A Study of Adaptation in Social Life (1985) by Thomas Seeley, on pages 81-83, estimates on honey and pollen consumption by honey bee hives are reported. Based on studies performed by this lab, Seeley estimates that in Connecticut, unmanaged honey bee hives averaging 30,000 adult bees in size have a yearly food consumption of 20 kg of pollen and 60 kg of honey. This is further separated into consumption values of 25 kg of honey in the winter and 35 kg of honey in the summer. This summer period is the 22 weeks between late April and late September during which it is estimated that colonies consume a combined 55 kg of pollen and honey. Using these estimates, a honey bee colony would consume 35 kg of honey during the 22 week summer period or 1.59 kg of honey per week. This independent estimate of honey consumption is approximately half of that provided to the colonies during the clothianidin colony feeding study.

Seeley (1985)[2] cites additional work on colonies managed for honey production in Europe and North America with a honey consumption range of 60 to 80 kg of honey per year. Note that Seeley's own work with unmanaged colonies represents the low end of this estimate. Assuming a proportional increase, a colony at the highest end of this range would be expected to consume 2.12 kg of honey per week (1.59 kg/wk times the ratio of the 80 kg to 60 kg). This amount is also less than the weekly amount provided to colonies during the colony feeding study.

Honey bees are known to collect nectar for consumption and to store excess as honey. It is important to recognize that sucrose solution removed from a feeder does not equate solely to consumption during feeding intervals. The fact that in almost all cases the bees of a respective colony remove all of the sucrose solution from the feeder does not indicate that there was insufficient sucrose solution in the feeder to meet the dietary needs of the colony during that feeding interval. During the feeding period the hives increased in weight and the honey/nectar estimates from the colony condition assessments show increases in cell numbers. This would be expected if the dietary needs of the honey bees were being met during the exposure period. Note that the colonies are free foraging and are presumably also bringing in some quantity of nectar in addition to the treatment solutions. It would also be expected that some of the provided treatment solutions were being stored long term as honey which is supported by chemical analysis of hive matrices collected after the feeding portion of the exposure period was complete.

In the endpoint evaluations for the clothianidin and thiamethoxam colony feeding study DERS, the Agency analyzed adults, eggs, larvae, and pupae numbers individually and combined together. Similarly, bee bread and honey/nectar cells were analyzed similarly. The Consortium questions the biological significance of these analyses and their potential interpretations. Combining these endpoints and analyzing them both individually and together is not common in honey bee research or other regulatory tests. It is not clear what additional value these aggregate endpoints bring to the interpretation of the study results. How can a treatment result in a non-significant effect on honey or bee bread residue values separately, but still have a significant effect on the combination of the two combined? If there is an effect on the larvae numbers, then there would also most likely be an effect on total brood (an aggregate of eggs, larvae, and pupae) and on the total individuals (an aggregate of adults, eggs, larvae, and pupae). Because multiple significant differences are likely to be observed in this situation, it has the appearance of multiple endpoints being affected and should be interpreted with caution. In this scenario it is also likely that the pupal numbers would be affected as well. In both aggregates, the endpoints will be dominated by one of the endpoints. For the total individuals, capped brood numbers are substantially higher in control colonies than the other life stages. For food stores, honey/nectar cells predominant over bee bread cells. Any results will generally reflect the component that is most numerous; therefore it is unlikely that these aggregate analyses add increased interpretation of study results

4.0 Use of Bee Bread as an Exposure Route

In the Preliminary Pollinator Assessment for clothianidin and thiamethoxam, EPA proposed use of a new method, termed the "Bee Bread" method, for assessing risk at the colony level to honey bees. EPA

[2] Seeley TD (1985) Honeybee ecology. Princeton University Press, Princeton.

reasoned that a direct comparison can be made between residues measured in nectars of various crop and non-crop plants to colony-level endpoints based on concentrations in spiked sucrose solution fed to honey bees in colony feeding studies. However, honey bees are also exposed to pollen in their diet and the Agency has not had a method to compare residues in pollen to a colony-level endpoint derived from a study in which the colony was fed only spiked sucrose solution. The Agency noted that honey bees mix pollen with nectar to form what is known as "bee bread" that is stored in the hive and used as a food source especially by nurse bees that tend to the queen and bee brood. EPA further reasoned that residues in bee bread could be estimated by weighting the residues in pollen and nectar (from crops) based on their relative contributions in bee bread. A review of the relevant literature (details in Appendix A of the Agency's assessment) led the Agency to estimate bee bread as being comprised on a dry weight basis by 55% pollen (P_{pollen}) and 45% nectar (P_{nectar}). On a fresh weight basis, pollen was further assumed to have 90% dry content and 10% water content, whereas nectar was assumed on average to have 30% dry (sugar) content and 70% water content. One unit of bee bread on a dry weight basis is therefore equal to 2.1 units on a fresh weight basis, as follows:

Bee Bread $_{\text{fresh weight}}$ **= (P_{pollen} / pollen dry wt fraction) + (P_{nectar} / nectar dry wt fraction)**

= (0.55 / 0.9) + (0.45 / 0.3) = 0.61 + 1.50 = 2.11

On a fresh weight basis, pollen comprises 0.61 / 2.11 = 0.29, or 29% of bee bread fresh weight,

and nectar comprises 1.50 / 2.11 = 0.71, or 71% of bee bread fresh weight.

To calculate the expected concentration in bee bread, one simply needs to combine the pollen and nectar residues which are typically reported on a fresh weight basis using these weighting factors.

Bee Bread residue = (Pollen residue x 0.29) + (Nectar residue x 0.71) ***Equation A***

Thus, if the concentration in both pollen and nectar was 10 ppb, the calculation would be

10 x 0.29 + 10 x 0.71 = 10 ppb.

This makes empirical sense, if the concentration of the two components is the same, the concentration of the mixture doesn't change.

If the concentration in pollen was 0 and the concentration in nectar was 20 ppb, the calculation is

0 x 0.29 + 20 x 0.71 = 14.2 ppb

However, EPA did not follow the above calculation formula (Equation A), but instead derived Equation 1 (reproduced below) which allows for combining residues of both clothianidin and thiamethoxam into clothianidin equivalents and uses a different fresh and dry weight conversion.

Equation 1

$$C_{bread-clothi} = \left[\frac{C_{pollen-thia} * 0.856 + C_{pollen-clothi}}{1 - 0.10} * 0.55 + \frac{C_{nectar-thia} * 0.856 + C_{nectar-clothi}}{1 - 0.70} * 0.45\right] * (1 - 0.25)$$

Where:

$C_{bread\text{-}clothi}$ is the concentration of clothianidin equivalents in bee bread (expressed as µg a.i./kg-ww);

$C_{pollen\text{-}thia}$ and $C_{pollen\text{-}clothi}$ are concentrations of thiamethoxam and clothianidin (expressed as µg a.i./kg-ww) measured in pollen at the same time point in samples collected from the same field;

$C_{nectar\text{-}thia}$ and $C_{nectar\text{-}clothi}$ are concentrations of thiamethoxam and clothianidin (expressed as µg a.i./kg-ww) measured in nectar at the same time point in samples collected from the same field.

If the scenario being considered involves only clothianidin residues, this equation reduces to

$$\mathbf{C_{bread} = [\ (C_{pollen} \times 0.55 / 0.9) + (C_{nectar} \times 0.45 / 0.3)\ ] \times 0.75}$$

The 0.75 multiplier at the end of the equation is supposed to convert from dry weight back to fresh weight, but the dry weight fractions of pollen (0.9) and nectar (0.3) are already present elsewhere in the equation and what is missing is the relative fresh weight fraction of each constituent material. It appears that something is amiss with this equation, because if you take the case where the concentration in both pollen and nectar is 10 ppb, Equation 1 calculates a concentration for the bee bread of 15.8 ppb, which is impossible. In fact, equation 1 always produces a value 1.58 times greater than does Equation A. Equation A was not presented by EPA in their assessment, but it is easily derived from the information presented in that assessment. Equation 1 appears to be invalid and needs to be replaced with Equation A.

While the above error in the calculation formula for residues in bee bread needs correction, there is a more fundamental flaw in the Bee Bread risk assessment method. As it is being applied, the Agency is assuming that the response of the test colonies in the colony feeding studies is being driven by the concentration present in the bee bread and ingestion of additional spiked sucrose solution produces no effect at all. Daily pollen and nectar intake rates for various castes of honey bees, as presented in the Agency's risk assessment guidance and the BeeREX model, indicate that all castes of adult bees ingest a higher proportion of nectar in their diet than pollen, typically an order of magnitude more nectar than pollen. The dose taken in by individual bees is therefore not just a function of the concentration in the bee bread. It is influenced to a greater extent by the concentration in nectar. Table 1 summarizes BeeREX model assumptions about pollen and nectar intake of the various castes of worker bees.

All pollen intake is assumed to come through ingesting bee bread. Bee bread is composed of 28.95% pollen. Therefore, in order to ingest 6.65 mg of pollen, workers that perform cell cleaning and capping would ingest 6.65 / 0.2895 = 22.97 mg of bee bread per day. This bee bread would consist of some nectar, and to ingest a total of 60 mg of nectar, these bees would need to ingest an additional 43.68 mg of nectar per day. Likewise, nurse bees that tend the queen and brood would ingest 33.16 mg of bee bread and 116.44 additional mg of nectar per day. As is evident from Table 1, other castes of worker bees ingest much less bee bread.

For example, what would be the total dose accumulated by these bees if their nectar food source was spiked with clothianidin and their pollen food coming in to the hive had no clothianidin in it? This question is directly relevant to interpreting the effects observed in the registrant-submitted colony-feeding studies, because this is precisely the scenario the bees experience in these studies. If the concentration in the sucrose solution fed to the bees was 20 ppb, which was the approximate no effect

concentration for clothianidin, the bee bread concentration would be 14.2 ppb (calculated using Equation A). The dose taken in by the various castes of worker bees would be as presented in Table 2.

Table 1. Daily intake of pollen, nectar and bee bread for the castes of adult worker honey bees. Estimates derived from the standard assumptions of EPA's BeeREX model.

	Pollen intake (mg per day)	Nectar intake (mg per day)	Intake of bee bread (mg)	Additional nectar intake (mg)
Worker (cell cleaning and capping)	6.65	60	22.97	43.68
Worker (brood and queen tending, nurse bees)	9.6	140	33.16	116.44
Worker (comb building, cleaning, food handling)	1.7	60	5.87	55.83
Worker (pollen forager)	0.041	43.5	0.14	43.4
Worker (nectar forager)	0.041	292	0.14	291.9
Worker (maintenance of hive in winter)	2	29	6.91	24.09

Table 2. Daily doses from the ingestion of bee bread and nectar for castes of adult worker honey bees. Calculations use the standard assumptions of EPA's BeeREX model and assumed source concentrations of 20 ppb and 0 ppb for nectar and pollen, respectively.

	Intake of bee bread (mg)	Additional nectar intake (mg)	Dose from bee bread (ng ai)	Additional dose from nectar (ng ai)	Total Dose (ng ai)	% of Total Dose from Bee Bread
Worker (cell cleaning and capping)	22.97	43.68	0.33	0.87	1.20	27.2%
Worker (brood and queen tending, nurse bees)	33.16	116.44	0.47	2.33	2.80	16.8%
Worker (comb building, cleaning, food handling)	5.87	55.83	0.08	1.12	1.20	6.9%
Worker (pollen forager)	0.14	43.40	0.002	0.868	0.870	0.23%
Worker (nectar forager)	0.14	291.9	0.002	5.838	5.840	0.03%
Worker (maintenance of hive in winter)	6.91	24.09	0.10	0.48	0.58	16.9%
						Average = 11.4%

The key result from Table 2 is that 0% to 27% of the daily dose taken in by adult worker bees is expected to come from ingestion of bee bread. The average for all castes of worker bees is 11% and for the castes of workers that remain in the hive (*i.e.*, non-foragers), the average is 17%. This means that more than 80% of the daily dose experienced by hive bees is expected to come from ingesting nectar/honey. These percentages, while calculated in Table 2 assuming source concentrations were 20 ppb for nectar and 0 ppb for pollen, are the same for any nectar dietary levels if the pollen concentration is 0. Thus, these percentages apply to all exposed colonies in the registrant-sponsored colony-feeding experiments.

Given that the vast majority of the dose ingested by the treatment groups in the colony feeding study did not come from ingestion of bee bread, it is not reasonable to assume, as the Agency did, that the response of these colonies was primarily related to the bee bread concentration. For example, one should not conclude that reduced colony strength measurements that were noted in the 40 ppb (nominal) sucrose-treatment group, the lowest concentration where adverse effects were observed, were the result of exposure to clothianidin concentrations in the bee bread that averaged 12 ppb. It is therefore inappropriate to use 12 ppb as a toxicity benchmark to compare with residue levels in plant pollen. The Agency's working hypothesis should be that effects are related to the total dose of clothianidin equivalents taken in by bees in the hive. The dose resulting from ingestion of pollen or bee bread is a minor component of the total dose. It is expected 80% or more of this dose comes from ingestion of nectar.

Empirical evidence was generated during the imidacloprid pilot studies in NC and MT that supports the conclusion that it is the concentration of chemical in the nectar food, not the pollen food of a bee colony, that drives the occurrence of adverse effects. In these pilot studies, colonies were fed dose levels of 0, 50 or 200 ppb either in artificial nectar, in artificial pollen, or at various combinations of these levels in both artificial nectar and pollen simultaneously. The response (reduction in colony strength measurements) observed in these test colonies was driven by the nectar concentration, not by the concentration in the artificial pollen.

In the pilot study conducted in North Carolina, colonies fed spiked sucrose showed clear reductions in the adult bee population (dashed lines in Figure 1) and amount of capped brood (dashed lines in Figure 2) that followed a concentration-response relationship, but colonies fed spiked pollen substitute showed no consistent differences between test levels for these same endpoints (solid lines in Figure 1 and Figure 2).

In the pilot study conducted in Montana (Figure 3, Figure 4), simultaneous exposure of colonies to 200 ppb in artificial pollen diet and 50 ppb in artificial nectar diet produced the same response as colonies fed 50 ppb in both artificial pollen and nectar diets. Feeding colonies 200 ppb in both pollen and nectar diets produced roughly the same result as feeding colonies 200 ppb in the nectar and 50 ppb in pollen. Clearly, the magnitude of the response was driven by the concentration in the artificial nectar. This pattern of response makes sense when one considers the parameters of the BeeREX model that indicate that bees on average get a much greater dose from ingestion of residues in nectar than they do from ingestion of residues in pollen.

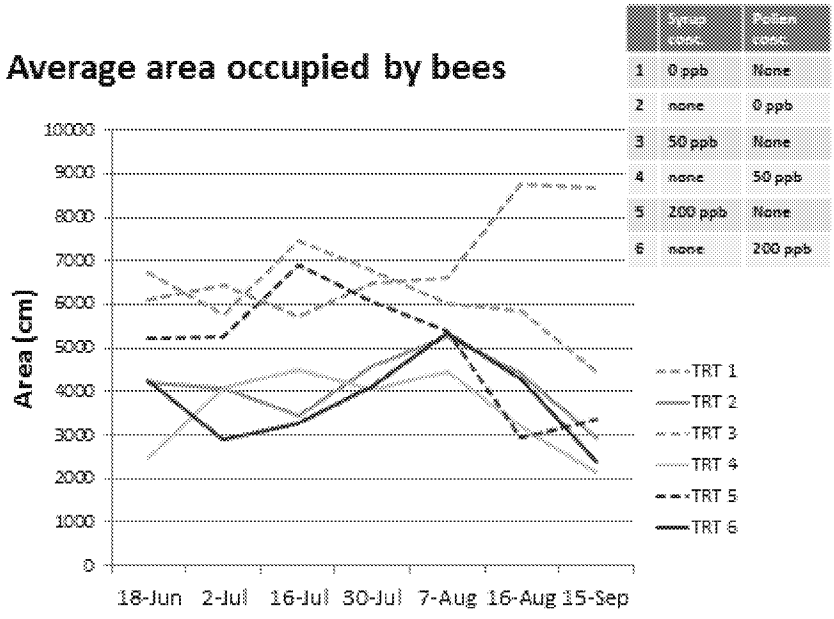

Figure 1. Colony strength (frame area occupied by bees) for colonies in North Carolina exposed to different concentrations of imidacloprid in either artificial nectar or pollen diets.

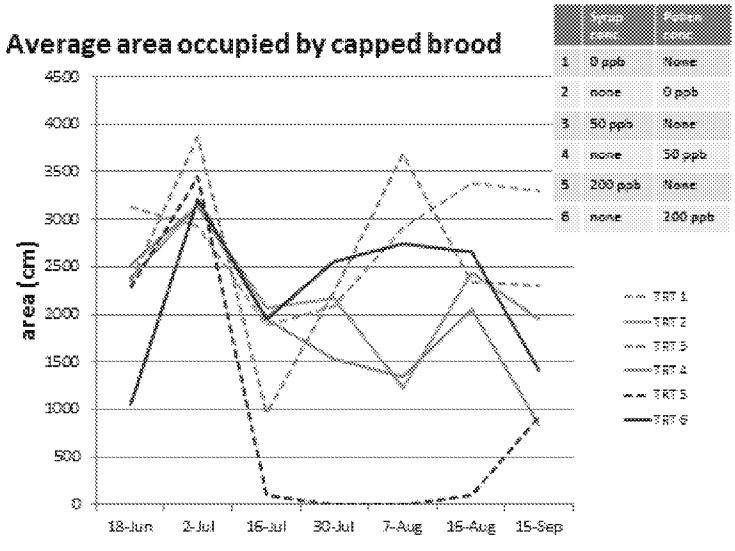

Figure 2. Colony strength (frame area occupied by capped brood) for colonies in North Carolina exposed to different concentrations of imidacloprid in either artificial nectar or pollen diets.

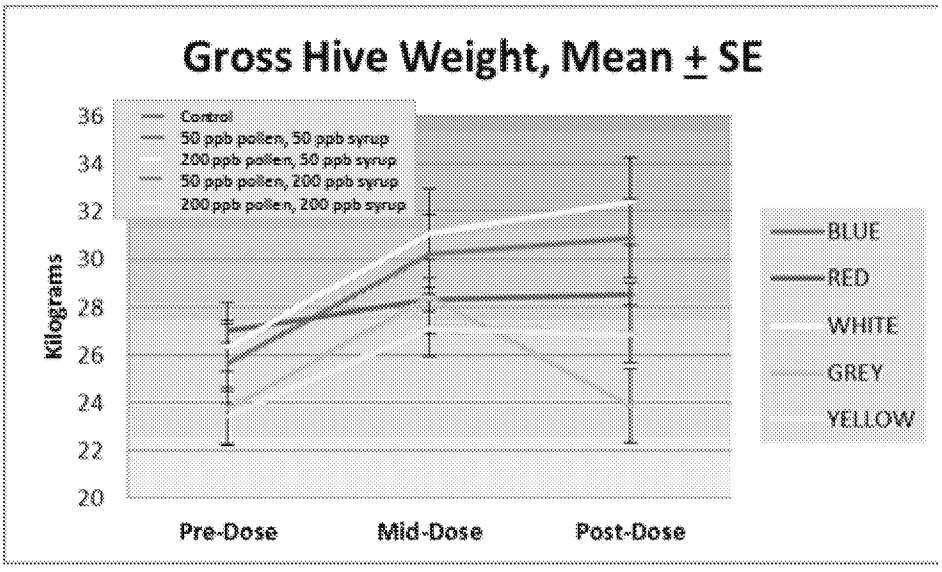

Figure 3. Weight of colonies in Montana exposed to various concentrations of imidacloprid in both artificial nectar and pollen diets.

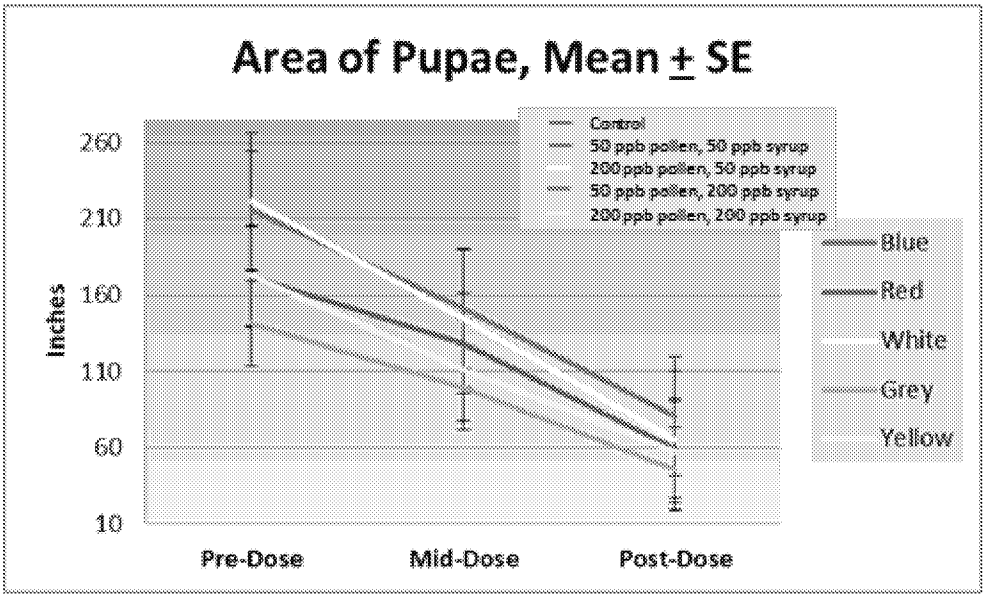

Figure 4. Colony strength (frame area occupied by capped brood) for colonies in Montana exposed to different concentrations of imidacloprid in either artificial nectar or pollen diets.

Given the results of the imidacloprid pilot study, the decision was taken to feed test colonies of the definitive study spiked sucrose solution only. This was a decision made in consultation with the EPA reviewer who agreed that the pilot studies demonstrated that nectar consumption was the driving factor in producing an effect. EPA approved the test protocol for this study, and for subsequent colony feeding studies with clothianidin, thiamethoxam and dinotefuran that also fed only spiked sucrose solution to the test colonies. So long as residues in pollen are of the same order of magnitude as residues in nectar, one would expect the response of colonies to be driven by the concentration in the nectar, and no specific assessment of risk to residues in pollen is likely needed except for instances in which the crop being treated is a significant source of pollen, but not nectar.

The colony feeding study by Dively *et al.* (2015)[3] adds additional support to the idea that the concentration causing effects is much higher when bees are exposed through residue in their pollen-based diet, in comparison to when exposed through sucrose-based diet. The Agency determine that the threshold effect level for bees fed artificial pollen patties spiked with imidacloprid was approximately 100 ppb, which is much higher than the 20-25 ppb no-effect level found in the various studies when colonies were fed spiked sucrose solution. As explained above, the finding that it takes a higher concentration in pollen to produce a colony-level effect is consistent with the BeeREX model assumptions about the relative contributions of pollen and nectar to the total diet of a colony.

In contrast, the studies of Sandrock *et al.* (2014)[4] and Williams *et al.* (2015)[5] might be seen as showing that colonies provided spiked artificial pollen experience effects at relatively low concentrations. However, the Williams study is not directly comparable to any of the other colony-feeding studies as it evaluated colonies set up for mass rearing of queens which involves very different colony management practices. The effect reported by Williams (queen failure) has not been found to occur in field studies (*e.g.*, Cutler *et al.* 2014[6], Rundloff *et al.* 2015[7]) of conventionally-managed colonies or by commercial beekeepers. The Williams study was also a small study (6 test colonies of which 3 received pesticide treatment) that employed pseudoreplication in the experimental design. It may therefore be dismissed as unsuitable for use in risk assessment. The Sandrock study reported a small difference in colony strength endpoints immediately after the exposure period which disappeared by the time the colonies were getting prepared for winter. Then there was a dramatic difference between treatment and control colonies the following summer for which there is no explanation from a toxicological perspective. For example, it is not clear if the bees were even still being exposed at the time these colony failures

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[3] Dively GP, Embrey MS, Kamel A, Hawthorne DJ and Pettis JS (2015) Assessment of chronic sublethal effects of imidacloprid on honey bee colony health. PLOS ONE | DOI:10.1371/journal.pone.0118748

[4] Sandrock C, Tanadini M, Tanadini LG, Fauser-Misslin A, Potts SG and P. Neumann (2014). Impact of Chronic Neonicotinoid Exposure on Honeybee Colony Performance and Queen Supersedure. PLoS ONE 9(8): e103592. doi:10.1371/journal.pone.0103592.

[5] Williams, G. R.; Troxler, A.; Retschnig, G.; Roth, K.; Yanez, O.; Shutler, D.; Neumann, P.; and L. Gauthier (2015) Neonicotinoid pesticides severely affect honey bee queens. Sci. Rep. 5, 14621; doi: 10.1038/srep14621.

[6] Cutler & Scott-Dupree (2014) A field study examining effects of exposure to clothianidin seed-treated corn on commercial bumble bee colonies. Ecotoxicology 23 (9), 1755-1763.

[7] Rundlöf M., Andersson G.K.S., Bommarco R., Fries I., Hederström V., Herbertsson L., Jonsson O., Klatt B.K., Pedersen T.R., Yourstone J., Smith H.G (2015) Seed coating with a neonicotinoid insecticide negatively affects wild bees. Nature 521, 77–80

occurred, or even at any time during their lives. A significant weakness of the Sandrock study is that only a single exposure level was tested and it is not clear if the results would be reproduced along a dose-response gradient. The demonstration of a robust dose-response relationship is a significant strength of the registrant-submitted colony feeding studies.

In conclusion, it is clear that the vast majority of pesticide exposure to a honey bee colony comes from ingestion of nectar and not pollen. Therefore, it is inappropriate to assume that response of honey bee colonies is a function of the residue levels in bee bread only. Based on dietary intake rates, one should expect a lower threshold concentration for adverse effects if a honey bee colony is fed spiked sucrose solution in comparison to if it is fed spiked pollen. This result has been confirmed in colony feeding experiments with imidacloprid. Thus, empirical data show a marked contrast to the Bee Bread method which predicts a lower threshold for adverse effects when colonies are exposed to residues in pollen.

Recommended path forward for Tier II risk assessment:

At the Tier II level, the Agency wants to compare residue measurements made in bee food items for various product use scenarios to a colony-level toxicity benchmark. The existing colony-feeding study provides the necessary colony-level toxicity benchmark. This benchmark is the highest no adverse effect concentration in artificial nectar fed over a chronic exposure period. While this benchmark is established in a test in which the test colonies were fed only spiked sucrose solution, it can be related to the concentration measured in the overall diet of a colony after the pollen and nectar components of the diet are expressed as nectar equivalents and then added together.

The food intake parameters of the BeeREX model suggest the contribution to the overall dose averages approximately five to ten times greater for nectar exposure in comparison to pollen exposure for worker bees performing various tasks inside the hive. As a conservative approach appropriate for use in a Tier II assessment, it is suggested that the concentration in pollen can be converted to nectar equivalents by dividing by a factor of 5. Then the total dietary concentration is determined as follows:

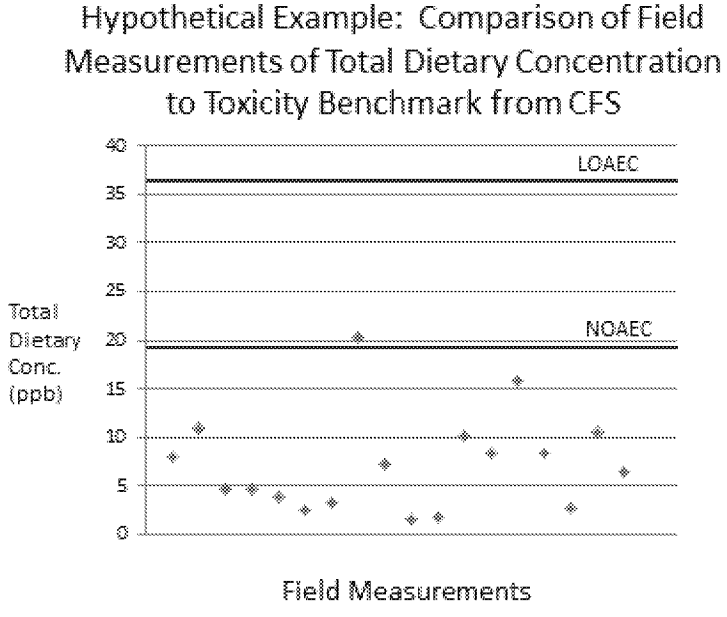
$$\textbf{Total Dietary Concentration}_{\text{nectar equivalents}} = \textbf{Nectar concentration} + (\textbf{Pollen concentration} / 5)$$

The field measurements of the total dietary concentration may then be compared to the colony feeding study NOAEC and LOAEC values and a determination made whether the risk is acceptable based on the frequency of exceedance of these benchmarks.

For example, assume the colony feeding study determined the NOAEC to be 19 ppb and a LOAEC of 36 ppb based on feeding the colony only spiked sucrose solution, and the following data were obtained from field measurements. (Note: these are not real data, but have been made up to illustrate the method.) The total dietary concentration in nectar equivalents has been calculated per the formula above and presented in the far right column.

Site	Replicate	Nectar conc (ppb)	Pollen conc (ppb)	Total diet concentration in nectar equivalents (ppb)
1	1	6	10	8.0
	2	8	15	11.0
	3	2	13	4.6
2	1	3	8	4.6
	2	2	9	3.8
	3	2	3	2.6
3	1	2	6	3.2
	2	15	27	20.4
	3	6	6	7.2
4	1	1	3	1.6
	2	1	4	1.8
	3	8	11	10.2
5	1	7	7	8.4
	2	12	19	15.8
	3	6	12	8.4
6	1	2	4	2.8
	2	7	18	10.6
	3	4	12	6.4

After calculating the total dietary concentration, these field measurements may be compared to the effects benchmarks derived from the colony feeding study, per the figure below.



In this hypothetical case, one of eighteen field measurements exceeds the colony feeding study NOAEC value, and just by a small amount. None of the field measurements approaches or exceeds the colony

feeding study LOAEC. The overall conclusion would be that the risk to honey bee colonies is low for this particular use pattern.

5.0 Risk Evaluation to non-*Apis* Bees

In the preliminary pollinator assessment for the nitroguanidine neonicotinoid insecticides, a comprehensive weight of evidence analysis to evaluate potential risk to non-*Apis* bees was presented. This analysis considered several lines of evidence, including (i) the USDA crop attractiveness guidance for bumble bees (*Bombus* spp.) and solitary bees (*Osmia* spp. and *M. rotundata*), (ii) consideration of the relative exposure of *Apis* and non-*Apis* bees, (iii) comparison of residue concentrations to Tier I non-*Apis* bee toxicity data, (iv) comparison of sensitivities of *Apis* and non-*Apis* bees in Tier I laboratory studies, (v) review of available colony level effects studies for non-*Apis* bees, and (vi) identification of potential uncertainties.

The Consortium supports the comprehensive EPA analysis. The ability to reliably determine a no effect-concentration in non-*Apis* species or estimate their exposure levels in a quantitative basis is still limited. Work in both aspects of the risk assessment is progressing, but further research is recognized to be needed. In the interim, the Consortium agrees with the EPA that there is sufficient body of evidence for neonicotinoids suggesting that honey bees are reasonable surrogates.

The following comments are offered on certain aspects of the Agency's review:

Tier I Toxicity Data

It is recognized in the EPA review that standardized toxicity protocols with non-*Apis* species are not currently available, which explains the variation in test designs and results in the existing studies. As with honey bees, the Consortium considers necessary the use of validated protocols for testing non-*Apis* bees in order to ensure robust risk assessments. By using validated tests, it is assumed that the methods are sufficiently reliable, repeatable and reproducible by different laboratories. Given that semi-field and field tests are required when concerns have been identified at lower tiers, there is also a need to develop suitable and validated test protocols for higher-tier tests.

Comparison of Sensitivities of Apis and non-Apis in Tier I laboratory studies

There is an increasing interest in the toxicity of pesticides to a wider range of pollinators and in factors which can be used for extrapolating pesticide toxicity from honey bees to non-*Apis* species to ensure risk assessments are protective (Thompson 2016)[8]. These differences may be due to specific detoxification capacities, but also to differences in body size as indicated in the EPA review. Mass-specific metabolic rates increase with decreasing body size. Thus, the Consortium agrees with EPA in using toxicity endpoints adjusted for bodyweight in their comparison between *Apis* and non-*Apis* data. Future research is still needed to properly address differences in sensitivity to pesticides across bee species, including honey bees, bumblebees, solitary bees and stingless bees. In particular average bodyweight for non-*Apis* species should be agreed by experts.

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[8] Thompson, H. (2016). Extrapolation of acute toxicity across bee species, Integrated Environmental Assessment and Management 12(4), 622–626. http://doi.org/10.1002/ieam.1737.

Nonetheless, considering all the existing data, the Consortium agrees with the Agency's overall conclusion that non-*Apis* bees show comparable sensitivities to *Apis*, suggesting that honey bees are reasonable surrogates.

Relative Exposure of Apis and non-Apis Bees

Screening-level acute contact EEC values for bumble bees were proposed by EPA. These EECs are based on an assumed bodyweight of 0.256 g for *Bombus* from a registrant-study (MRID 4957070). As recognized in the EPA review, the average bodyweight for bumble bees may vary from study to study or between species, introducing uncertainty in this exposure assessment. Agreement on average bodyweight for bumble bees should be reached by experts to ensure more robust exposure estimates.

Screening-level acute oral EEC values for bumble bees were also proposed by EPA based on food consumption rates from open literature. Likewise for solitary bees, there is uncertainty associated with these consumption rates according to non-*Apis* bee scientists. In addition, exposure to non-*Apis* bees could be also correlated with body weight because both food intake (metabolic rate) and surface area affect oral and contact exposure respectively (Thompson 2016). It is already recognized for bees that there is a relationship between body size and foraging distance: larger bees forage disproportionately farther than smaller bees (Greenleaf et al. 2007)[9]. The Consortium believes that agreement by non-*Apis* bee experts is needed to introduce the appropriate food intake rates in pesticide risk assessments.

Lack of basic knowledge of non-*Apis* bee exposure scenarios has been one of the biggest challenges in determining whether honey bees are good surrogates. A workshop was organized in Washington D.C. on 10th-12th January 2017 as a result of a tripartite effort between regulatory agencies, academia and industry. Forty bee researchers and risk assessors from ten different countries gathered to specifically discuss the latest state of science on pesticides exposure to non-*Apis* bees and to determine how well the honey bee exposure estimates used by different Regulatory Agencies may or may not cover other bee species. After three days of discussions there was a general consensus that the current honey bee exposure assessment paradigm is highly conservative. However, several data gaps were identified that hindered the full quantification of exposure to non-*Apis* bees, especially when these bees are exposed via nesting materials such as soil (e.g., blue orchard bees; *Osmia* spp., alkali bees; *Nomia* spp.), leaves (e.g., leafcutter bees, *Megachile rotundata*) or a combination of soil and leaves (e.g., stingless bees; *Meliponini* spp.). Basic conceptual models and preliminary exposure equations were discussed that will help in future to address the quantification of these alternative exposure routes and, subsequently, will allow comparison with honey bee exposure estimates. The Consortium recommends continuous efforts on the development of these areas in order to advance in risk assessment to bee pollinators.

Review of Available Colony Level Effects Studies for non-Apis Bees in Open Literature

Several studies in the open literature suggest the occurrence of effects of clothianidin and/or thiamethoxam for bumble bees and solitary bees (*Osmia* spp) when exposed to similar or levels below those reported in the registrant colony feeding studies (MRID 49836101 and 49757201), but not in all cases. The Consortium agrees with the Agency that all these studies have test design limitations and cannot be used in risk assessment.

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[9] Greenleaf SS, Williams NM, Winfree R, Kremen C (2007) Bee foraging ranges and their relationship to body size. Oecologia 153:589–596.

The following comments are offered on several papers reviewed by the EPA in which clothianidin, thiamethoxam or imidacloprid were tested. There were not references concerning available non-*Apis* studies in the dinotefuran assessment. In addition, other published studies are referred to where they provide information relevant to this assessment.

i. Tier II Studies

In the 11-week greenhouse study (Scholer & Krischik (2014)[10] caged queen right colonies of *Bombus impatiens* were fed treatments of 0, 10, 20, 50 and 100 ppb clothianidin or imidacloprid in 50% sugar syrup. These are nominal concentrations and the values given in the EPA review do not reflect the mean measured values given in the paper. The largest impact for both neonicotinoids starting at 20 ppb was the statistically significant reductions in queen survival, worker movement, colony consumption, and colony weight. It was concluded that bumblebees feeding on imidacloprid and clothianidin at 20 ppb can cause changes in behavior (reduced worker movement, consumption, wax pot production, and nectar storage) that result in detrimental effects on colonies (queen survival and colony weight). However, as previously noted, these dose levels are considerably in excess of those found in field crops, even before taking into account the worst-case nature of the study, with lack of choice. The study by Fauser-Misslin et al. (2014)[11] looked at the possible interaction between neonicotinoids and pathogens and so the comment in the EPA review that the statistical analysis only looked at the combined effect simply reflects the objective of the study. They showed that chronic dietary exposure from an early stage of *Bombus terrestris* colony development to doses of thiamethoxam and clothianidin that "could be encountered in the field" resulted in a significant interaction between neonicotinoid exposure and parasite infection on mother queen survival. However, the colonies were fed both treated pollen and nectar substitute for a prolonged period, from the 10-worker stage to the senescent phase. Thus, while this study demonstrates that effects of clothianidin treatments can be detected it involves use of exposure regimes that are unrealistic in relation to normal field conditions.

In their study on solitary bees, Sandrock et al. (2013, not 2014a as given in the EPA review)[12] investigated the influence of field-realistic trace residues of the routinely used neonicotinoid insecticides thiamethoxam and clothianidin in nectar substitutes on the entire life-time fitness performance of the red mason bee *Osmia bicornis*. They showed that chronic, dietary neonicotinoid exposure did have detrimental effects. Thus, while neonicotinoids did not affect adult bee mortality, monitoring of fully controlled experimental populations revealed that sublethal exposure resulted in almost 50% reduced total offspring production and a significantly male-biased offspring sex ratio. It was concluded that to fully mitigate long-term impacts on pollinator population dynamics, present pesticide risk assessments need to be expanded to include whole life-cycle fitness estimates, as demonstrated in the present study using *O. bicornis* as a model. However, the bees were fed a mixture of thiamethoxam and clothianidin at 'field realistic' concentrations of 2.87 and 0.45 ng/g, respectively, using artificial flowers in flight cages for up to 40 days. This represents an unrealistically severe exposure and the effects of clothianidin and

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[10] Scholer J, Krischik V (2014) Chronic Exposure of Imidacloprid and Clothianidin Reduce Queen Survival, Foraging, and Nectar Storing in Colonies of Bombus impatiens. PLoS ONE 9(3): e91573.

[11] Fauser-Misslin A, Sadd BM, Neumann P and Sandrock C (2014) Influence of combined pesticide and parasite exposure on bumblebee colony traits in the laboratory. J Appl Ecol 51:450-459.

[12] Sandrock, C., L. G. Tanadini, J. S. Pettis, J. C. Biesmeijer, S. G. Potts, P. Neumann (2013) Sublethal neonicotinoid insecticide exposure reduces solitary bee reproductive success. Agricultural and Forest Entomology, 16: 119-128.

thiamethoxam under field conditions is addressed under Section 4.3 (Tier III) of the EPA review. Also, this study was designed to reflect field exposure of honey bees to neonicotinoid treatments and so cannot be used for the clothianidin or thiamethoxam risk assessment, as the treatment involved a mixture of both compounds and therefore any observed effects cannot be attributed specifically to either compound.

ii. Tier III Studies

In the case of bumble bees, Rundlöf et al. (2015, not 2014 as given in the EPA review) reported a negative effect on hive weights and reproduction measured as the number of new queens. In another study by Sterk et al. (2016)[13], colony development in terms of hive weight and the number of workers showed a typical course with no statistically significant differences being found between the sites. In addition, the reproductive output (young queens and queen brood) cells were comparatively high and not negatively affected by the exposure to treated oilseed rape. In the Rundlöf et al. (2015) study, non-standardized hives were used and the colonies were terminated at the first sight of emerging queens, which can be an incomplete way to measure reproductive performance.

Rundlöf et al. (2015) also found that nesting activities of *Osmia bicornis* completely ceased on fields adjacent to oilseed rape fields treated with a clothianidin containing formulation. However, these results are based on a very low sample size (27 cocoons were placed at each field) and there was a low average emergence from these cocoons compared to the rate achieved in the latter study. As a result, there was a low number of exposed individuals and in such circumstances non-treatment related effects, e.g. predation and adverse weather conditions can have a significant influence on the data. In the study reported in Schmuck and Lewis (2016)[14], where 1500 cocoons were placed at each of three locations in each test area, overall reproductive output of *Osmia bicornis* was high at both sites and parasitization rates remained low (less than 3%) and within the natural range. A statistically significant higher percentage of oilseed rape pollen was collected by mason bees in the reference site compared to the test site, which may have been due to variability in oilseed rape pollen availability or the availability of alternative forage. Also, significantly more males than females were produced at the test site but the number of females did not differ significantly between test and reference sites and their mating system requires more males to be present. Accordingly, no evidence for an adverse effect on the overall fitness of populations of red mason bees was found.

Perhaps the most important difference between the two studies, common to all the species tested, is that different crops were used. Rundlöf et al. (2015) used spring oilseed rape, which is sown in spring and flowers in the same year as it is planted, while winter oilseed rape (as used in Schmuck and Lewis study) flowers in the year after it has been sown. Winter oilseed rape is commercially a much more important crop in Europe and provides an important resource for pollinating insects due to its prolonged flowering period. Spring planted oil-seed rape is a minor crop in Europe, but a major crop in North America where it is referred to a canola. However, the seed-treatment rate that Rundlöf et al. studied was 250% greater than the maximum rate labeled for use in North America. As the time between

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[13] Sterk G, Peters B, Gao Z, Zumkier U (2016) Large-scale monitoring of effects of clothianidin dressed oilseed rape seeds on pollinating insects in Northern Germany: Effects on large earth bumble bees (*Bombus terrestris*). Ecotoxicology. doi:10.1007/s10646-016-1730-y

[14] R. Schmuck, G. Lewis (2016) Review of field and monitoring studies investigating the role of nitro-substituted neonicotinoid insecticides in the reported losses of honey bee colonies (*Apis mellifera*). Ecotoxicology; doi: 10.1007/s10646-016-1734-7.

sowing and flowering for spring oilseed rape is shorter than in winter oilseed rape, higher residue levels can be expected for spring oilseed rape if the seeds are treated at the same rate. However, with the lower rate labeled for use in North America, residue levels in canola are typically in the same low range reported by Schmuck and Lewis, and much lower than those reported by Rundlöf. The study reported by Schmuck and Lewis is considered to be representative of the majority of oilseed rape cultivation areas in Europe and reflective of the risk to both managed and wild bee species from the use of clothianidin seed-treated winter oilseed rape.

In another study conducted by Nicholls et al. (2017)[15], the effect of exposure of *Osmia bicornis* larvae to a range of field-realistic concentrations (0 to 10 ppb) was examined. Six `trap nests', consisting of cardboard tubes housed in a waterproof shelter, were positioned a few meters above ground in an orchard. Nests were placed out in early April 2016, and each contained a release tube seeded with 12 female and 10 male cocoons. Cocoons were checked for viability and sex prior to release. From mid-May onwards, once bees had emerged, tubes were checked daily for the presence of eggs. Eggs plus pollen provisions were removed from the nests and added to individual polystyrene nest blocks. Nest blocks were then returned to the laboratory and weighed before being placed into a dark incubator. Bees were assigned to one of four treatments; 0 ppb (control), 1 ppb, 3 ppb or 10 ppb clothianidin, with exposure being made via their pollen provisions. The results showed no effect on larval development time, overwintering survival or adult weight. Flow-through respirometry was used to test for latent effects of larval exposure on adult physiological function: no effect of larval clothianidin exposure was observed. These results confirm those in the study reported by Schmuck and Lewis (2016).

Szabo et al. (2012)[16] considered the decline of several North American bumblebee species in the light of possible pathogen spillover from commercial bumblebees. They used a large dataset of bumblebee occurrence records and agricultural census data and found support for the pathogen spillover hypothesis for two species but not for the near disappearance of the previously widespread *Bombus affinis*. In addition, they showed that pesticide use and habitat loss are unlikely to be major causes of decline for any of the *Bombus* species examined. This latter conclusion was criticized by Stevens and Jenkins (2012)[17] who suggested that the paper relied on out of date data and was missing a key class of pesticides, the neonicotinoids. Without data that include these widely used systemic insecticides, it was considered that the authors could not reliably assert that pesticides have not played a role in bumblebee declines across North America. While the authors agreed that better tracking of neonicotinoid input from various treatments is needed to assess their wildlife impacts (Colla et al., 2013)[18] it was pointed out that the available data indicate that neonicotinoid use does not explain broad-scale declines among the three eastern North American bumblebee species studied. This is supported by recent evidence that these species began exhibiting declines prior to the registration and widespread use of neonicotinoids in North America.

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[15] E. Nicholls, R. Fowler, J.E. Niven, J.D. Gilbert, D. Goulson (2017) Larval exposure to field-realistic concentrations of clothianidin has no effect on development rate, over-winter survival or adult metabolic rate in a solitary bee, *Osmia bicornis*. PeerJ 5:e3417; DOI 10.7717/peerj.3417.

[16] N.D. Szabo, S.R. Colla, D.L. Wagner, L.F. Gall, J.T. Kerr (2012) Do pathogen spillover, pesticide use, or habitat loss explain recent North American bumblebee declines? Conservation Letters 5, 232-239.

[17] S.M. Stevens, P.T. Jenkins (2012) Pesticide impacts on bumblebee decline: a missing piece. Conservation Letters 6, 213-214.

[18] S.R. Colla, N.D. Szabo, D.L. Wagner, L.F. Gall, J.T. Kerr (2013) Response to Stevens and Jenkins pesticide impacts on bumblebees: a missing piece. Conservation Letters 6 (3), 215-216.

The paper by Cutler et al. (2014) referred to in the section on *Bombus* (page 180 in the EPA clothianidin and thiamethoxam review) is actually a study examining the effects of exposure to clothianidin seed-treated canola on honey bee colony health, development and overwintering success. The relevant paper described with respect to effects on bumble bees is actually Cutler and Scott-Dupree (2014)[19]. In this study clothianidin was detected (0.1-0.8 ng/g) in pollen collected from all conventional fields, but was not detected in pollen from organic fields. Corn pollen was only rarely collected from bumble bee foragers and the vast majority of pollen was from wild plants around the corn fields. All hives appeared healthy and neonicotinoid seed treatments had no effect on any hive endpoints measured, except the number of workers, where significantly fewer workers were removed from hives placed next to conventional fields compared to organic. In terms of the most important parameter for bumble bees, queen production (both number and weight), was unaffected by clothianidin treated seed and was actually higher (by >25%) compared to organic. Consequently, it was concluded that the results suggest that exposure during pollen shed to corn grown from neonicotinoid-treated seed poses low risk to *B. impatiens*.

6.0 Off-field Drift Assessment

To assess the potential risk to bees exposed to drift in off-field habitat, EPA used the BeeREX model (v.1.0) to determine risk quotients (RQs) for acute contact and oral and chronic oral exposure routes based on foliar application rates. The level of concern (LOC), which is 0.4 for acute and 1.0 for chronic, divided by the RQ determined the drift fractions that would be acceptable such that the RQ was less than the LOC for acute and chronic exposure. The drift fraction was then used with the AgDRIFT® model (v. 2.1.1) to estimate the distance at which acceptable drift deposition would occur for ground and aerial applications. The distance required for the drift fraction to be low enough such that the RQ no longer exceeded the acute or chronic LOC for acute contact and oral as well as chronic oral scenarios was determined. The Neonicotinoid Consortium considers this approach to be overly conservative for assessing risk to bees that may be exposed to pesticides from drift off-field. This critique is particularly important given that this method was not a component of the bee risk assessment process that was vetted with the Scientific Advisory Panel (SAP) and is not part of the formal bee risk assessment guidance (US EPA, PMRA, CDPR 2014).

The Neonicotinoid Consortium believe that any spray buffers that might be recommended based on off-field assessments are not necessary based on current pollinator protection goals and given the fact that label language is already in place that prohibits drift to flowering crops and weeds with instructions to minimize the availability of blooming plants prior to application. Thus prior to and during application, measures need to be taken that will minimize any off-field exposure and potential risk to bees foraging in off-field field habitats.

The method used by EPA is overly conservative regarding exposure. The off-field risk assessment method uses conservative default values for droplet size, boom height and wind speed percentiles as inputs to the AgDRIFT model. These inputs can be refined based on label specific language. Additional aspects, pointed out by the Agency in the assessment, that likely lead to overestimation of exposure are

[19] Cutler & Scott-Dupree (2014) A field study examining effects of exposure to clothianidin seed-treated corn on commercial bumble bee colonies. Ecotoxicology 23 (9), 1755-1763.

that the model assumes there is no interception by the crop canopy and that winds are unidirectional and constant to the off-field area.

Off-field spray drift is predominantly composed of the smallest droplet sizes (driftable fines) that do not deposit on plant structures (i.e. leaves, stems, flowers) in the same fashion as a direct, saturating overspray due to the nature of atmospheric mechanisms impacting the dispersion of airborne particles and their interaction with solid surfaces. As the spray cloud moves away from the sprayed area, the droplet size distribution shifts toward a smaller liquid volume comprising a greater proportion of driftable fines (less than ~100 µm). The smaller droplets generally lack sufficient momentum to penetrate the boundary layer of air surrounding plant foliage, thus producing lower actual exposures than predicted by deposition curves. This may vary across different plant structures based on differences in capture efficiency resulting from difference in surface roughness, turbulent mixing and other factors, but in all cases, is expected to render lower exposure values than predicted by a deposition-based model like AgDRIFT.

The method is overly conservative regarding effects. Tier I laboratory-based effects endpoints (acute oral and contact LD50; chronic oral NOED) are used to determine acceptable drift deposition distances. A more realistic approach would be to use No Observed Adverse Effect Rates (NOAER) from Tier II semi-field tunnel studies if available, and compare those rates to the AgDRIFT deposition curve to identify distances equivalent to the NOAER that would protect the colony. However, comparison to tunnel study NOEARs can still be considered highly conservative as the application of the pesticide in a semi-field study is made directly to the crop while bees are foraging, thus bees can receive a direct application from the spray and higher contact exposure from the leaves and flowers due to the direct application rather than deposition of driftable fines on the plants.

The proposed EPA method incorporates both contact and oral exposure routes over both acute and chronic exposure durations. From a dietary standpoint, the amount of drift that could potentially land on pollen and/or nectar is likely much lower compared to what could potentially land on leaf material. Furthermore, off-field habitat immediately adjacent to a crop where a foliar application is made, and the proportion of the habitat where spray drift actually deposits on the plants, taking into consideration plant interception, is likely small, and would not be a significant portion of the overall feeding range of the colony foragers. Any pollen and nectar collected from adjacent areas would not result in a significant proportion of the colony provisions suggesting that the oral route of exposure is not likely to be an important component in assessing off-field risk to bees. The chronic endpoint is also based on a continuous oral exposure even though degradation, based on available pollen and nectar residue studies, can be substantial. As acknowledged by the Agency on page 205 of the thiamethoxam/clothianidin assessment, "...chronic exceedances are based on repeated exposures at the same concentration and does not take into account the degradation of the chemical" and, therefore, chronic exposure is likely overestimated. Given the number of overly conservative assumptions concerning both the route and duration of exposure for off-field drift to bees, the Neonicotinoid Consortium believe that the acute and chronic dietary component should be removed and if any off-field assessment is needed, the focus should be on assessing the potential risk to bees from acute contact exposure to spray drift.

The Neonicotinoid Consortium recommendation is that the Agency should consider label language that prohibits drift to off-field plants when they are in bloom. Furthermore, the Agency should consider

refinements to the AgDRIFT model when supported by label language to provide the best estimate of potential exposure considering the drift deposition estimates are highly conservative. If available, NOAERs from semi-field tunnels should be used to compare rates to the refined AgDRIFT deposition curve to identify distances appropriate for protecting honey bee colonies. If such higher-tier data are not available, the acute contact LD50 should be used in conjunction with BeeREX and AgDRIFT to determine acceptable distances such that acute LOCs are not exceeded. Acute oral and chronic oral risk components are not necessary as the potential area of forage that would receive drift deposition would be small compared to the forage range of honey bees and drift deposition onto pollen and nectar would be low such that potential risk from oral exposure would be minimal.

7.0 Use of Pollen and Nectar Residue Data in the Exposure Assessment

The Neonicotinoid Consortium has submitted to the Agency a very extensive crop residue data set (nectar and pollen) developed to derive bee exposure estimates. The registrants recognize the use of all available data for measured residues in nectar and pollen from treated crops in the risk assessment by EPA and consider that this is highly appropriate. However, it is noted that currently there is no guidance on the collection of such data and its use in the risk assessment process. Initially the refined Tier I assessment is based on these worst-case default residue values. Subsequently, in the refined Tier I assessment and at Tier II, all available registrant-residue data are introduced i.e. both from additional trials (where available) and considering changes over time. It is considered that the EPA's approach to deriving estimated environmental concentrations (EECs) from the submitted data is overly conservative as it uses the overall maximum values and the maximum mean values for individual sites even when these are likely outliers and do not represent the majority of the data.

The Consortium believes that a better use of all the available residue data could be made to derive a more realistic assessment of the exposure levels. It is suggested that for the acute risk, 90th percentile residue levels can be derived from these data sets while for the chronic risk, overall mean values from all sites can be used. This approach has been adopted in the EU, e.g. for the risk assessment of birds and wild mammals (EFSA 2009)[20]. In this case, standardized exposure values are calculated for various focal species, crop and growth stage combinations using all available residue data in each case. For the acute risk, 90th percentile residue levels are derived from these data sets while for the chronic risk, mean values are used. Given the relatively large variation found in field-collected residue data, risk assessments are always going to be subject to the inherent variability of such data and the use of potentially unrepresentative worst-case values. Using the distribution of the available residue data is both more realistic in relation to field conditions and can be considered sufficiently protective, if used appropriately.

8.0 Seed Treatment Dust Stewardship

As mentioned in the clothianidin and thiamethoxam assessment, exposure of bees via drift of abraded seed coat dust is considered a route of concern, but "the Agency is working with different stakeholders to identify best management practices and to promote technology-based solutions that reduce this

[20] FSA (2009) Risk assessment for birds and mammals. EFSA Journal 2009; 7(12): 1438.

potential route of exposure." Minimizing dust drift resulting from planting treated seed is among the highest priorities for the member companies of the Neonicotinoid Consortium. Examples of our extensive efforts in this regard include developing and optimizing new seed treatment formulations and tank-mix recipes to minimize dust abrasion through the use of new and improved dust reducing agents and polymers. In addition, we provide extensive applicator training to our seed treatment customers, including seed companies and treaters, on how to properly handle and apply our products during the seed treatment process. Further, we offer assistance to these same customers at their treater sites when questions and issues arise to ensure our seed treatment products are properly applied. Through our sales and seed advisor staff, we educate growers on the best way to handle and plant treated seed to minimize dust abrasion and dust-off at planting as well as disposing leftover treated seed. Some registrants of Neonicotinoids conduct extensive research in seed treatment application technology to determine the best seed processing steps (i.e., cleaning seed before treatment) all the way to evaluating and optimizing droplet/particle sizes during seed treatment application to ensure our products stay on the seed while handling and planting to minimize dust-off.